

## Fungal and marine shell fouling in desalination plant equipment

Mohamed O. Saeed, Ghazzai F. Al-Otaibi and M. I. Mohamed Ershath

### ABSTRACT

The Saudi Arabian Saline Water Conversion Corporation (SWCC) aims to maintain an uninterrupted desalinated water output and has tasked its Desalination Technologies Research Institute (DTRI) with trouble-shooting operational problems and unusual events faced by its desalination plants. Three events were reported and investigated by DTRI. Two were found to involve fungal fouling, and one was found to involve fouling by marine shells. One case of fungal fouling involved a new seawater reverse osmosis membrane and the plant was advised to review the handling and storage practice of membranes. The other case involved product water hoses and manifested itself in the form of black slimy deposits arising from dense fungal growth. The fungus originated from new product hoses and was eliminated by shock-dosing replacement hoses with chlorine. The marine shell fouling involved a feed water line of a combined power/desalination plant. Chlorine, hydrochloric acid, ethylenediaminetetraacetic acid, and fresh water were used to assess their ability to control marine shell fouling in laboratory experiments, with varying results. Since high doses of chlorine were not effective in controlling marine shell fouling, the practice of continuous chlorination should be abandoned in favor of an alternative chlorination regimen, e.g., pulse chlorination.

**Key words** | chemical agents, chlorination, desalination equipment, fungal fouling, marine shell fouling

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### INTRODUCTION

Saudi Arabia produces 5.6 Mm<sup>3</sup> of desalinated seawater per day; this amounts to approximately 22% of the total world production and 45.5% of production in the Gulf region. One-third of this quantity is produced by seawater reverse osmosis (SWRO) plants. Biofouling of reverse osmosis (RO) membranes is a major cause of operational problems in SWRO plants in all regions of the world. SWRO plants in Saudi Arabia are more prone to biofouling because they are situated in a hot climate, which is more

conducive to microbial growth and attachment (Saeed *et al.* 2000; Jamaluddin *et al.* 2001; Hassan 2002).

To avoid contamination from sources other than feed water, careful handling and proper storage of desalination equipment must be ensured. It was noticed that a new membrane from a stack of membranes in a storeroom at an SWRO plant on the southern Red Sea coast of Saudi Arabia was leaking its preservative fluid due to a small rupture in its protective case. It was decided not to use the membrane out of fear of airborne contamination. The membrane was removed and investigated for microbial contamination. This plant, with a production capacity of 2,275 m<sup>3</sup>/d, was commissioned in 1983.

A case of slimy black deposits on the inner side of the product water hoses was reported from an SWRO plant

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along the mid-northern section of the Red Sea coast. The plant was commissioned in 1986 with a production capacity of 4,400 m<sup>3</sup>/d. The plant uses copper sulfate as a biocide and had no history of membrane fouling. Several years after commissioning, black deposits were observed on the inside of most of the product water hoses of one of the six membrane trains of the plant. The remaining trains were free of the deposit. Product water hoses in the affected train were replaced, but the product hoses of the other trains were not replaced. Microscopic examination of wet mounts prepared from the black deposits revealed dense fungal growth. Multiple microbiological isolations and membrane autopsies were carried out to determine the nature and origin of this fungal fouling. Autopsy was performed on the lead and end membrane elements of a pressure vessel of the affected train, as well as on the product hose itself. Autopsied membranes, as well as product hoses, were subjected to various analyses, including visual inspection, inspection by scanning electron microscopy (SEM) and microbiological isolation to identify the root cause of the problem.

Macrofouling (commonly known as marine shell fouling) is a common problem for the equipment and structures (such as intake structures) of seawater desalination plants because of their proximity to the sea. Laudable efforts have been made around the world to understand the phenomenon of macrofouling and evolve strategies for its prevention and control (WHOI 1952; Crisp 1973). At times, the accumulation is so dense that it calls for cleaning methods to be investigated (Abdul Azis *et al.* 2003). Chlorine is normally the chemical agent of choice for controlling biofouling in water systems because of its broad-spectrum effectiveness and low cost (Rajagopal *et al.* 1995). Other chemicals used to clean marine shell-covered surfaces include hydrochloric acid (HCl) and ethylenediaminetetraacetic acid (EDTA). A solution of 50% HCl in water dissolved barnacle shells in less than 2 min (Dolez & Love 2002). EDTA is usually used as a chelating and buffering agent (Moropoulou & Kefallonitou 2002).

A study was carried out to investigate the best control method for marine shell growth inside feed lines of a power/multi-stage flash (MSF) desalination plant. The plant, which produces 1.40 Mm<sup>3</sup> of desalinated water per day and generates 1200 MW of electricity, is located on the Gulf coast of Saudi Arabia and was commissioned in 1983.

Approximately 12.5 Mm<sup>3</sup> of feed water per day is pumped from coastal waters for cooling and desalination purposes.

The aims of the present study are: (1) to provide results of the analysis of three biofouling incidences involving desalination equipment, particularly the very rarely reported fungal fouling and (2) to encourage the practice of attention to detail and proper inspection by desalination plant operators.

## EXPERIMENTAL METHODS

### Membranes

The membrane at the southern Red Sea coast plant consists of cellulose triacetate and has a hollow fine fiber (HFF) configuration. The membrane at the mid-northern Red Sea coast plant is a thin film composite polyamide of spiral wound (SW) configuration.

### Microbiological examination of membranes

The HFF membrane was autopsied employing aseptic techniques. Samples of the membrane fibers were obtained from the top, middle and innermost fiber layers. Wet mounts of the fibers were made in sterile physiologic saline and examined under a compound microscope. Because fungal hyphae were observed in the wet mounts, fibers were tested for fungal growth. Fibers were aseptically excised and spread on Sabouraud's Dextrose Agar, a medium enhancing fungal growth, in standard size Petri dishes. The agar dishes were incubated in a thermostatically controlled high-low temperature incubator at 25 °C and observed for any fungal growth for a period of 7 days.

The autopsy was also performed on the SW membrane in the system with fouled product hoses. Samples of the membrane sheet and the spacers of lead and end elements were aseptically excised for analysis. Excised pieces were each suspended in triplicate in 10 ml of sterile physiologic saline and vortexed to remove the attached biofilm. Biofilm remaining after vortex mixing was carefully removed by scraping with a sterile plastic spatula. Following appropriate sample dilution, pour plate seeding was employed in Nutrient and Sabouraud's Dextrose Agar media to obtain heterotrophic plate counts of bacteria/fungi.

### Microbial examination of feed water

To trace the origin of the contamination of the product water hoses, water samples for microbial analysis were taken from the intake area in the open sea and from four locations along the pretreatment line: after coagulant dosing, after the micron cartridge filter, after the clear well, and from the brine reject.

The sampling points were first cleaned with a brush and flushed for 10 min; they were then covered for 10 min with paper towels and soaked with household bleach. The sampling points were then flushed for a further 5 min, after which water samples were taken in sterile plastic sampling bags and analyzed immediately. The bacteria/fungi were counted following the incubation of samples at a temperature of 25 °C. The samples were first mixed well on a vortex mixer, after which pour plate counts were carried out in Sabouraud's Dextrose Agar supplemented with 2% salt and in Marine Agar (Difco 2216).

### Microbial analysis of fouled hoses

Wet mounts from the black deposit as well as from new hoses stored in the plant (as replacements for used ones) were prepared and examined under a compound photomicroscope. The hoses were also photographed. Sabouraud's Dextrose Agar (used as is or supplemented with 2% salt) and Marine Agar were used for fungal/bacterial isolation from the fouled and new hoses. This was accomplished by inoculating the two media with cotton swabs from the hoses.

### Scanning electron microscopic analysis

SEM analysis was carried out on the fibers of the fouled membrane and on fouled product water hoses. Cut pieces of unused replacement hoses were also subjected to SEM analysis in order to trace the origin of the fouling deposit.

Specimens for SEM analysis were air-dried and placed on a brass plug using double-sided carbon (conductive) tape. They were then coated with 30 nm of gold, using a JEOL-1100E ion-sputtering device. The specimens were placed in the JEOL model JSM-5300LV SEM and were observed using a 20 kV accelerating voltage at a working distance of 10 mm.

Loss of mass on ignition was used to estimate the percentage of organic matter in the membrane deposits and consequently the extent of membrane fouling. This was accomplished by ashing a dried and weighed sample of the membrane deposits in a crucible at 550 °C for 1 h.

### Marine shell fouling

Representatives of different marine shellfish taxa were collected live from broken feed lines (Figure 1) and from rocks in the intake bay of the power/MSF plant. The shells were barnacles, mussels (bivalve molluscs) and gastropods (monovalve molluscs), with mussels dominating at about 60% of the population. Specimens were subjected to chemical agents in the laboratory so as to devise a chemical cleaning method. The HCl, EDTA, HCL + EDTA, chlorine, and distilled water were tested as potential control agents for marine shell fouling.

Shellfish were distributed in glass beakers (1 l) containing test chemicals dissolved in aerated filtered (0.45 µm pore membrane filter) seawater. The impact of osmotic shock on shellfish was tested by subjecting them to distilled water, with filtered seawater used as control. Marine shells were exposed to the following combinations of HCl and EDTA: 5% HCl (stock HCl has 1.18 g/ml specific gravity and 37% concentration), 5% HCl + 5% EDTA (stock



**Figure 1** | Marine shells (s) in a feed water line of an MSF desalination plant on the Gulf coast.

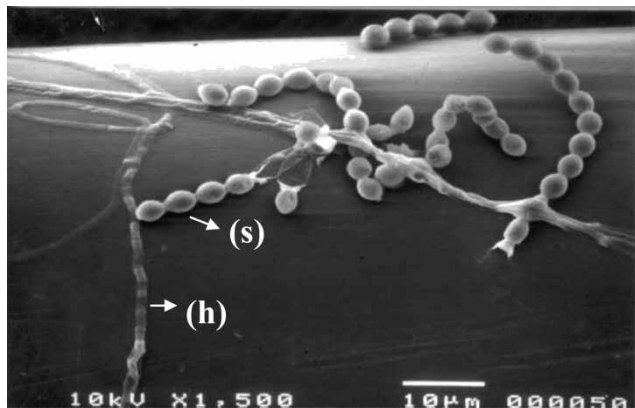
EDTA is 99.5% minimum assay), 5% HCl + 1% EDTA, 5% EDTA, and 1% EDTA. Chlorine concentrations were prepared from household bleach and included 0.3, 0.5, 1.0, and 5.0 mg/l. Each beaker received 12 animals (four from each of the three taxa) and tests were replicated thrice. Tests were performed at a room temperature of 23 °C.

The animals were observed under test conditions continuously during the day and then left overnight for a total of 24 h.

## RESULTS AND DISCUSSION

### Fouling of membrane

The HFF membrane from the SWRO plant on the southern Red Sea coast was found to be heavily infected with fungi. The fungal isolate spread into the deeper membrane layers



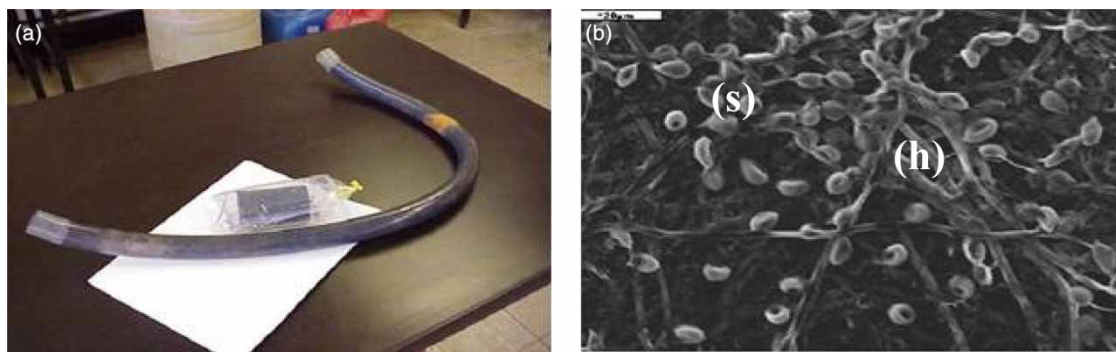
**Figure 2** | SEM of a single HFF of an SWRO membrane showing fungal hyphae (h) and spores (s).

and was readily isolated on the growth medium tested. Fungal hyphae were also readily apparent in wet mounts. SEM examination revealed fungal hyphae undergoing virulent sporulation, indicating a shortage of nutrients (Figure 2). The fungal fouling can be attributed to airborne spores, due to the ruptured protective case of the membrane and leaking of preservative fluid. The fouled membrane was discarded rather than used for fear of maloperation and spreading of the fouling agent to the whole installation. The plant was advised to review the handling and storage procedure of membranes.

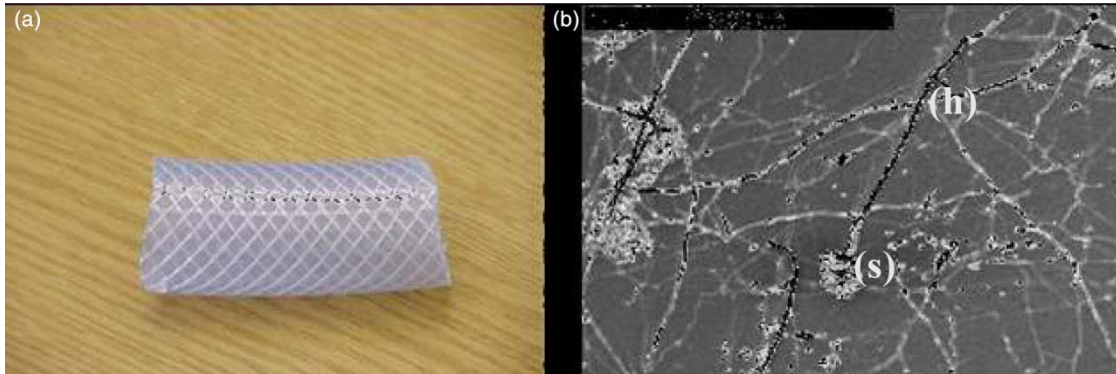
### Fouling of product water hoses

Wet mounts of the black deposit from the product water hoses at the SWRO plant in the mid-northern section of the Red Sea coast revealed a dense fungal contaminant (Figure 3). On agar media, the fungal colonies were white with fluffy hairy margins. The isolate was closely related to the genus *Aspergillus*. This isolate was also present on a sample piece from the new hose reel that was kept in the plant store for replacements (Figure 4).

Analysis of biofilm formation on membranes from the plant with the fouled product hoses showed biofilm bacterial density (total heterotrophic count) in the order of  $10^4$  CFU/cm<sup>2</sup>. No fungi were present on the membranes, indicating that membranes are not the source of fungal growth in product water hoses. These results suggest limited biofilm formation on the membranes (Jamaluddin *et al.* 2001) as fouled membranes normally contain an average density in the order of  $10^6$  CFU/cm<sup>2</sup> bacteria and up to  $10^3$  CFU/cm<sup>2</sup> fungi (Baker & Dudley 1998). The loss on



**Figure 3** | (a) Photograph of a fouled product hose showing internal fungal growth in the form of black slimy deposit and (b) SEM of the deposit showing fungal hyphae (h) and spores (s).



**Figure 4** | (a) Excised piece of a new product water replacement hose showing lack of growth by the naked eye and (b) SEM of the internal surface showing fungal hyphae (h) and spores (s).

ignition, which indicates primary organic matter, was about 50%. In fouled membranes, loss of ignition is more than 50% (Baker & Dudley 1998) or more than 70% (Flemming 1993). The loss on ignition results suggesting limited fouling are consistent with the results of the microbiological analyses. Visual inspection showed the membrane covered with a reddish-brown slimy deposit throughout its leaves (Figure 5). The deposit was iron from the coagulant ferric chloride.

Water samples taken from the intake area in the open sea and from four locations along the pretreatment line (after coagulant dosing, after the micron cartridge filter, after the clear well, and from the brine reject) showed no fungal growth.

Therefore, the results of the microbiological analyses of membranes and water samples are physical evidence that the fungal growth on the product water connection hoses

did not originate from source seawater. It should also be noted that the contaminating fungus was growing in hoses containing the product, which is fresh water. This also indicates that the fungus is not likely to have originated from the sea. The fouling is likely to have stemmed from the presence of fungus on new product hoses, which could not be observed by the naked eye and is revealed by SEM (Figure 4). The presence of the fungus on the new product water hoses suggests that the product hose replacement led to fungal growth. The black fungal growth was only observed in the membrane racks in which product water hoses had been replaced. The product hoses in other racks were free of any deposits. The problem was alleviated by replacing fouled product hoses by new hoses that were soaked overnight in a solution of 5% household bleach. As a precaution, the plant was advised to soak new replacement product hoses in a concentrated hypochlorite solution before use.



**Figure 5** | Autopsied SW membrane associated with a fouled product hose.

### Marine shell fouling

Toxicity tests on marine shellfish showed that all were killed by 5% HCl solution, with completely dissolved barnacle and bivalve shells, and partially dissolved gastropod shells. The same result was obtained for the solution of 5% HCl and 5% EDTA. The 5% HCl and 1% EDTA solution also resulted in 100% mortality, with dissolved barnacle shells, partially dissolved bivalve shells, but unaffected gastropod shells. With 5% EDTA, animals were killed with only a very light reaction on the shells. The 1% EDTA solution had no effect on either survival or shell integrity (Table 1).

**Table 1** | Effect of HCl<sup>a</sup> and EDTA<sup>b</sup> separate or in combination on marine shells

S. No.	Solution	pH <sup>c</sup>	Shells tested	Remarks
1	5% HCl	0.5	1. Barnacles 2. Bivalves 3. Gastropods	Barnacles and bivalves killed and shells completely dissolved, gastropods killed and shells partially dissolved, appreciable gas generated.
2	5% HCl + 5% EDTA	0.75	1. Barnacles 2. Bivalves 3. Gastropods	Barnacles and bivalves killed and shells completely dissolved, gastropods killed and shells partially dissolved, appreciable gas generated with appreciable precipitate.
3	5% HCl + 1% EDTA	0.93	1. Barnacles 2. Bivalves 3. Gastropods	Barnacles and bivalves killed, barnacle shells dissolved, bivalves shells partially dissolved, gastropod killed with intact shells.
4	5% EDTA	4.80	1. Barnacles 2. Bivalves 3. Gastropods	Barnacles and bivalves killed, slight reaction on shells with light gas production but no dissolution, gastropod killed but shells intact.
5	1% EDTA	5.30	1. Barnacles 2. Bivalves 3. Gastropods	No effect

<sup>a</sup>HCl used as is at 37% concentration.

<sup>b</sup>Ethylenediaminetetraacetic acid.

<sup>c</sup>pH measured after overnight incubation of solution with shells.

Although HCl proved to be an effective control and cleaning agent of marine shells, there is a drawback to its use: appreciable quantities of acid are required in large feed water-lines. The pH of 5% HCl solutions is less than 1.0, which is corrosive for valves in feed lines. Furthermore, an appreciable generation of carbon dioxide gas occurs from the acid's reaction with shells, which are composed primarily of calcium carbonate. It is estimated that 614,000 l of gas could be generated in a header based on the density of attached shells. The accumulation of gas in closed headers can lead to an explosion unless some kind of venting is provided.

The results of exposure to different free chlorine concentrations indicate animals to be very tolerant of this disinfection agent. Chlorine was not toxic to animals at concentrations up to 1.0 mg/l. At a concentration of 5.0 mg/l, mortality was only about 40%. Initially, the animals became listless and lost their footing. Mortality occurred within the first 40 min of exposure. However, the surviving shells gradually regained their motility and became reattached. Chlorination is the method most often applied to combat macrofouling because of its proven efficacy and low cost. However, the two commonly used dosing regimens are not ideal. Intermittent (shock) dosing (e.g., one hour per day) is only effective against microfouling (slime-forming bacteria) and not against marine shells

because they close their shells and/or stop feeding during the dosing period and recover easily when dosing stops. Shock chlorination consists of mixing sufficient chlorine-based chemical with water to create a solution of free chlorine concentration of approximately 15–300 mg/l, depending on pH and exposure time (Neb Guide 2007; Dixon *et al.* 2012). Alternatively, under low-level continuous dosing bivalves can close their shells for long periods, causing the chlorination to take several months to become effective (Dar Tec Engineering Consultants 2017). Continuous low dose chlorination was not found effective to deter barnacle larval settlement and metamorphosis in a coastal power station (Venkatnarayanan *et al.* 2016).

Clearly, continuous dosing of chlorine that is generated on-line (electrochemical generation) from seawater is not an ideal disinfection method. The plants should either revert to intermittent shock dosing at chlorine concentrations of >5.0 mg/l or seek alternative chlorination or disinfection methods. The present on-line chlorine generation system cannot provide shock-dosing concentrations. Also, high chlorine dosing comes with a high cost and serious environmental concerns. In both continuous and intermittent chlorination modes, chlorine is dosed even if there are no fouling organisms because these organisms flourish during certain seasons, resulting in wasted chlorine.

Pulse chlorination is an attractive alternative to continuous and intermittent chlorination modes (Dar Tec Engineering Consultants 2017). This is a technology based on short successive pulses of chlorine. Shelled animals open and close their valves at regular intervals. These intervals can be measured and a chlorine pulse initiated when the valves open. Pulse chlorination can reduce costs by 50% compared with other chlorination modes and has a reduced environmental impact due to reduced overall chlorine use (Dar Tec Engineering Consultants 2017). The concerned plant was advised to test pulse chlorination and is preparing a pilot test of this technology.

Marine shells exposed to distilled water, all died within 20 min of exposure. After 10 min exposure to distilled water, the shells lost their footing on walls of the test beakers. They settled to the bottom and became listless and died within the next 10 min. Therefore, distilled water in an attractive control and cleaning alternative. If the feed lines were designed in such a way to allow for their isolation and treatment with product water (filling and draining), then this method would be the most cost effective and environmentally friendly.

## CONCLUSIONS

1. RO membranes with defective protective envelopes are subject to airborne contamination and should be discarded without ever being used.
2. Black deposits inside SWRO membrane product water hoses were found to be due to fungal growth. The fungal growth could be traced to contaminated new hoses and not to source seawater or membrane fouling.
3. Continuous chlorination is not an ideal control method for marine shells in intake pipes of desalination and power plants.
4. Distilled water was found to be effective in killing marine shelled animals.

## RECOMMENDATIONS

1. Attention to detail and proper inspection of the equipment of desalination plants should be practiced.

2. Replacement SWRO membrane product hoses should be soaked in 5% household bleach before installation.
3. Continuous chlorination should be abandoned in favor of alternative chlorination methods.
4. Product water can be used to control marine shell growth in intake pipes.

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